8.5–1.3 and 5.5–1.7, respectively. The decrease by treatment was statistically significant at the 0.05 confidence level.

Discussion. The inhibition of ADP-induced platelet aggregation by addition of *Aspergillus* enzyme to PRP in vitro was reported by BYGDEMAN[8] and DE NICOLA et al.[9]. The latter investigators also, observed this action of the enzyme in vivo by i.v. infusion[9].

Our results show that proteolytic enzyme counteracted the aggregation of unwashed as well as washed platelets, induced by ADP or thrombin.

Hyperacid ACD[7] used as an anticoagulant, exclusion of red cells by respinning and washing of platelets with 5.4 m*M* sodium citrate[10], promoted the preparation of washed platelets without spontaneous aggregation maintaining their proper aggregability.

Although the deprivation of platelet sensitivity to ADP by washing had been reported[11], restoration of aggregability by inclusion of calcium, magnesium, glucose and albumin in the suspending fluid was described by ARDLIE et al.[12]. But, because of unsuitability of providing extraneous protein as a component of platelet suspending solution in the present study, thrombin was chosen as an aggregating agent instead of ADP. The enzymatically treated platelets were rewashed prior to mixing with thrombin to avoid further enzymatic action of thrombin.

The effectiveness of protease on washed platelets would at least partially suggest that the inhibitory effect of proteolytic enzymes on platelet aggregation could be an effect of enzymes on platelet membrane.

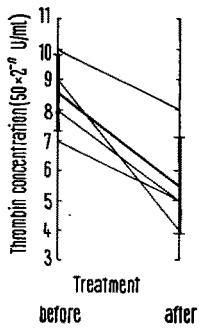

Fig. 3. Effect of protease on thrombin-induced aggregation of washed platelets.

Zusammenfassung. Behandlung ungewaschener und gewaschener Thrombozyten mit proteolytischen Enzymen (Protease, Bromelain und Ficin) bewirkt eine Abnahme der Aggregationsfähigkeit von Thrombozyten mit ADP oder Thrombin.

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Influences of Nystagmic and Spontaneous Oculomotor Activity on Superior Colliculus Neurons in Curarized Cat

The influence of eye movement on neurons in primary visual structures of cat has recently been studied by several authors. Modifications of neuronal activity related to eye movements have been described at the level of lateral geniculate body[1] and tectum opticum[2]. The aim of the present study was to investigate the influence of nystagmic and spontaneous oculomotor activity on superior colliculus (SC) neurons in absence of actual eye movements, thus avoiding possible retinal image shift or proprioceptive input.

Material and methods. Cats were prepared under ether anesthesia. A high cervical transsection was performed ('encéphale isolé'); expiratory CO_2 and body temperature were monitored. After recovering from the surgical procedure, animals usually exhibited a satisfactory amount of spontaneous and pursuit eye movement. Nystagmus was induced by polarization of labyrinths. Horizontal eye movements were recorded by electrooculography. A concentric bipolar macroelectrode was then introduced stereotactically in or in the close neighborhood of nucleus abducens. Correlation between eye movement and synchronous oculomotor discharges was established for each animal. Glass microelectrodes were introduced through the cortex into the SC and the animal curarized with Flaxédil. Visual stimuli consisted in switching on and off a diffuse light, or hand-moving bright or dark objects. In absence of these stimuli, the animal was facing a structureless background (photopic conditions). Both eyes were left open. Microelectrode tip, at the end of a penetration, was localized by electrophoresis of pontamine[3]. The brain was perfused with formalin, frozen and cut serially. Blue spots were used to reconstruct the electrode track and the location of the units within the collicular layers.

Results and discussion. The influence of nystagmic oculomotor activity on spontaneous firing pattern of 82 collicular neurons was studied. Their visual characteristics were determined by methods described above: 46 units (56%) were thus found to be unresponsive to visual stimulation.

The effects of nystagmic oculomotor activity may be roughly divided into 2 categories. Spontaneous activity of 9 collicular neurons (11%; group I) was phasically modified in synchrony with nystagmic oculomotor discharges recorded at the level of nucleus abducens (Figure, I). 2 cells were inhibited, the others were activated. 3 units were found to be visually driven. A second set of 7 neurons (9%; group II) showed 'tonic' modifications of activity during sequences of nystagmic oculomotor discharges. These effects are rather complex and, most frequently, consist of an increase of spike frequency (Figure, II). 2 of these units were sensitive to movement of a visual stimulus, others could not be driven by visual stimulation.

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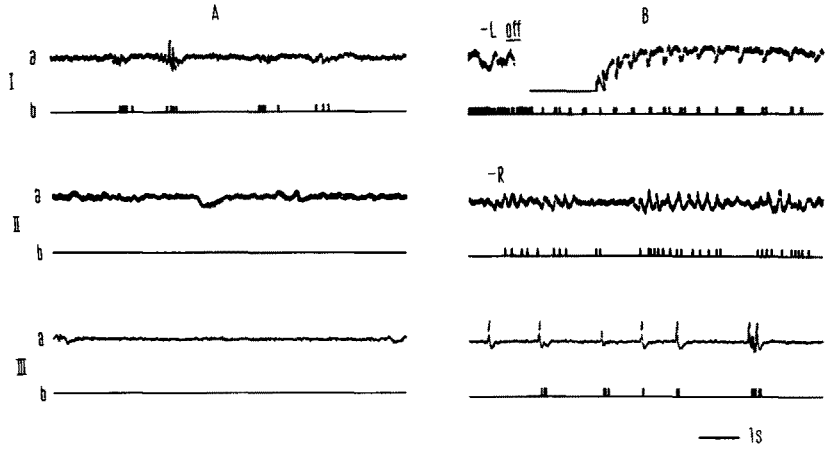

Illustration of the 3 groups of collicular neurons (see text). Group I. a) Nucleus abducens macropotentials, b) neuronal activity (spikes equalized by a Schmitt trigger), A) neuronal discharges synchronous with spontaneous oculomotor activity (OMA), B) same neuron showing phasic discharges time-locked with nystagmic OMA at the end of cathodal polarization of left labyrinth (off). Note activation during polarization.
Group II. a) and b) as in I, A) spontaneous oculomotor activity, no neuronal discharge, B) same cell showing tonic discharges during bursts of nystagmic OMA elicited by right cathodal polarization.
Group III. A) No spontaneous OMA, no neuronal discharge, B) neuronal discharges correlated with spontaneous OMA.

Whenever animals presented a satisfactory spontaneous oculomotor activity, its influence was analyzed. 14 neurons (group III) showed modifications of activity time-locked with such spontaneous discharges. These phasic changes of firing pattern consited of an excitation; 1 unit however was synchronously inhibited (Figure, III). 5 units in this group responded to stationary (on-off) or, in most cases, moving visual stimuli; the other cells could not be driven by visual stimuli. 4 of these nonvisual units were located in stratum periventriculare. Several neurons of this group also presented phasic modifications during nystagmic oculomotor discharges and are also included in the first group (Table).

Phase relationship between oculomotor discharges and phasic effects was measured. The burst of spikes or the inhibition takes place at a time interval ranging from a few ms before the initial peak of the abducens wave up to 300 ms after this peak. Phase relationship is in some cases quite constant; in other cases, specially during nystagmic activity, it may vary slightly from one wave to the other. The macropotentials, at the level of nucleus abducens, were themselves recorded 10 to 50 ms before the onset of the spontaneous or nystagmic saccade. All the effects related to oculomotor activity, specially during nystagmus, show a remarkable 'lability' in time. For the same cell, they may fluctuate from one oculomotor wave to the other and even disappear from a nystagmic sequence to the other. It is to be noted that, in our experiments, animals are fully awake during nystagmic tests. For some neurons, effects depend on the side of the polarized labyrinth. Finally, all the analyzed neurons are located in the various layers of the SC and the underlying periaqueductal gray. No correlation was found between type of effect and localization of neurons.

These observations show that visual as well as non visual cells in the SC receive information about oculomotor output, though no eye movement actually occurs. Curarization makes it possible to eliminate extraocular proprioceptive afferences which are known to reach deeper layers of SC[4]. Moreover, any visual stimulation due to image shift on the retina is avoided. Recording of nucleus abducens discharges, though relatively less accurate than electrooculography, allows us to keep a reliable cue of oculomotor orders. Direction of nystagmus may even be specified by observation of the shape of these waves. An interesting observation is that some neurons are identically influenced by spontaneous as well as by nystagmic oculomotor activity. According to studies of LORENTE DE NO[5] and SZENTAGOTHAI[6] on the vestibulo-ocular reflex arc, the only known site common to spontaneous and nystagmic oculomotor pathways lies at the level of brainstem oculomotor nuclei. This argument was already developed by ZUBER and STARK[7] for the saccadic suppression phenomenon, which is related to other observed visual oculomotor interactions, themselves embodied in the concept of 'corollary discharge'[8]. The influence of oculomotor activity evidenced in this study on visual but also on nonvisual neurons spread over all collicular strata might provide other experimental data in favour of the 'efference copy' postulated in von HOLST and MITTELSTAEDT[9] 'Reafferenzprinzip'.

Résumé. L'influence de l'activité oculomotrice sur les neurones colliculaires du chat «encéphale isolé» curarisé peut se diviser en 3 catégories: 1. décharges ou inhibitions synchrones à l'activité oculomotrice spontanée ou 2. nystagmique; 3. modifications toniques de l'activité spontanée durant des périodes de nystagmus.

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Classification of SC neurons influenced by oculomotor activity

Group I	Phasically influenced units during nystagmus	– visual	(A) 3
		– non-visual	(B) 6
Group II	Tonically influenced units during nystagmus	– visual	(C) 2
		– non-visual	(D) 5
Group III	Units influenced by spontaneous oculomotor activity	– visual	(E) 5
		– non-visual	(F) 9
	$A \cap E = 2$		
	$B \cap F = 1$		

Figures correspond to number of units. Note overlap between group I and III.

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